

# Physiological, Volatile, and SEM Surface Effects Resulting from Cutting and Dipping Treatments in Cantaloupe

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**Abstract:** Previous research examined sanitation treatments on cut cantaloupe tissue to deliver germicidal and food safety effects. However, an apparent compromise between volatile loss and treatment/sampling efficacy appeared. Subsequently, a physiological and volatile reassessment of thinly sliced tissue against cubes was performed in cantaloupe tissue. Thin sliced cantaloupe  $L^*$  decreased 27.5%, 40.5%, and 52.9% in 3, 2, and 1 mm thickness, respectively, compared with cut cubes after 3 d. Overall color (C) decreased in freshly prepared cubes (2.4%) and slices (14.4%) that were washed in cold water. Surface area per unit volume (SA : vol) in slices was 4.1 times greater than typical cubes, as reflected by substantial water loss (20.4%, 9.5%, and 6.7% in 1, 2 and 3-mm slices, respectively) after 1 d at 5 °C. Rinsing cubes and thin-slices with 5 °C deionized water resulted in roughly 15% soluble solids loss. SEM indicated 65.4% reduced cell size in 1-d old thin slices, evidenced by excessive cell damage and desiccation compared with stored fresh-cut cubes. In thin-sliced tissue exposed 15 min to an open atmosphere (mimic sanitation treatments), total esters decreased 92.8% and 95.8%, respectively, after 1 and 3 d storage at 5 °C. Washing tissue provided a boundary layer that reduced short-term ester losses in slices and cubes. Excessive cutting, sanitation treatment regimes, and storage can radically alter the desirable volatile profile of cut cantaloupe. Reduction of tissue size to maximize food-safety sanitation efficacy or delivering items to a niche market will need substantial work to engineer equipment and develop protocols to insure that product quality and volatiles are not compromised.

**Keywords:** aroma, cantaloupe (*Cucumis melo* L. Naudin), ester, flavor, scanning electron microscopy (SEM), soluble solids, water loss

**Practical Application:** We have demonstrated that cutting method and sampling protocol are critically important when using volatiles as a means by which to assess or interpret stress response and ascribe fresh-cut quality. Reduction of tissue size to maximize food-safety sanitation efficacy (for example, thin slices) will need substantial work to engineer equipment and design protocols to insure product quality and volatile profiles are not compromised.

## Introduction

It is well recognized that minimal processing and wounding have profound physiological effects on plant tissue (Toivonen and DeEll 2002; Soliva-Fortuny and Martín-Belloso 2003). Some consequences are considered desirable such as allium flavor release (Carson 1987),  $C_6$  and/or  $C_9$  aldehyde/alcohol generation in tomato (Riley and Thompson 1998), cucumber (Fleming and others 1968), bell peppers (Wu and Liou 1986), and melons (Beaulieu and Lea 2006; Schieberle and others 1990). However, most consequences are physiologically deleterious and have adverse effects regarding product integrity and quality, especially in

sensitive fruits (Karakurt and Huber 2007). The fresh-cut fruit market has not experienced phenomenal growth similar to its companion fresh-cut salad and vegetable market. This is predominately due to the inherently fragile nature of ripe fruit with removed epidermis, and also been attributed to likely flavor imbalance, and/or volatile loss (Beaulieu 2006a; 2007).

Cantaloupes pose a substantial food safety risk since their flesh is at neutral to basic pH, and the netting may harbor pathogens (Annous and others 2004). The FDA therefore mandates that fresh-cut cantaloupe be stored < 5 °C to slow down microbial growth, since it is considered a potentially hazardous food (PHF), posing food safety risk (U.S. FDA 2007). Subsequently, various sanitation treatments (electron beam, gamma irradiation, hot water, ozone, vacuum-steam) have been explored to reduce microbial loads on whole and cut cantaloupes to improve safety (Boynton and others 2005, 2006; Fan and others 2006, 2008; Selma and others 2007; Solomon and others 2006; Ukuku and others 2006; Wang and others 2006). Other studies have assessed the effects of ultraviolet radiation on thin-sliced cantaloupe (Beaulieu 2007; Lamikanra and others 2002). However, most published works testing sanitation treatments have understandably been focused on establishing

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effective treatment levels and microbial analysis, while volatile and sensory appraisals are generally lacking. Sensory evaluation rated 1 kGy e-beam irradiated fresh-cut cantaloupe highest in “sweetness,” “cantaloupe flavor intensity,” “juicy,” and lowest in “off-flavor” after long-term (17 d) storage at 3 °C (Boynton and others 2006). Irradiation causes softening (Boynton and others 2005) and subsequently, attributes like sweetness and juicy would be expected to “improve.” However, percent soluble solids or volatile data were not provided to deliver correlative interpretation. Physiological deterioration must be prevented with sanitation treatments, and it is likewise crucial that consumers accept the flavor and texture.

Volatile appraisal in most fresh-cut food safety sanitation treatments is lacking. Whether flavor and sensory attributes can be retained with oftentimes “abusive” sanitation protocols remains to be seen. UV-treatments applied to fresh-cut cantaloupe cubes indicated that fruity/melon and sweet aromatic notes were reduced; however, volatile analysis was not reported (Lamikanra and others 2005). Treatment-induced diffusional loss of flavor volatiles in thinly sliced UV-treated cantaloupe tissue was recently reported (Beaulieu 2007). Ester losses were presumed to be due to experimental procedures and off-gassing (Beaulieu 2007; Forney 2008). Subsequently, a physiological and volatile reassessment of thinly sliced tissue compared with cubes was performed to determine if food-safety-oriented sampling protocols compromise tissue integrity and subsequent post-cutting quality. Several sanitation treatments employ a bathing solution and dipping were evaluated. SEM and ester recovery were utilized to visualize surface effects and volatile changes in sliced compared with cubed tissue.

## Materials and Methods

### Plant material

A total of 4 western shipper orange-fleshed cantaloupe (*Cucumis melo* L. Naudin) cultivars were used. “Esteem” and “Sol Real” were grown under standard cultural practices with furrow irrigation in California, and 2 store-bought unknown cultivars, “cultivar 1” and “cultivar 2.” The store-bought cultivar 1 was judged very ripe ( $10.5\% \pm 0.1\%$  soluble solids,  $n = 10$ ) based on intense aroma and texture upon hand cutting, whereas the second cultivar was less ripe ( $10.2\% \pm 0.5\%$  soluble solids,  $n = 10$ ), lacking intense aroma and being more firm upon cutting. “Esteem” and “Sol Real” fruit were harvested, force air cooled, boxed with Styrofoam beads, air freighted overnight to the SRRC, and analyzed promptly upon receipt. They were commercially ripe (for example, for “Esteem,”  $\frac{3}{4}$ -slip,  $10.3\% \pm 0.6\%$  soluble solids,  $n = 18$ , and “Sol Real,”  $\frac{3}{4}$ -slip,  $10.9\% \pm 0.9\%$  soluble solids,  $n = 42$ ).

### Sample preparation

Whole cantaloupe were assessed for initial quality and sanitized in 100 ppm bleach (5.25% NaClO, pH approximately equal to 6.7), uniformly peeled on a Muro CP-44 melon peeler (Tokyo, Japan). To assess various physiological changes after washing, rinsing and mimicked sanitation treatments, uniform equatorial mesocarp tissue was used to prepare thinly sliced tissue (approximately 1, 2, and 3 mm; for example,  $1.94 \pm 0.08$  mm) on a Hobart 512TMS (Troy, Ohio, U.S.A.) or generally approximately 2.5 mm thickness (for example,  $2.67 \pm 0.11$  mm) via a hand-held mandolin kitchen slicer (Robinson Knife Co., Buffalo, N.Y., U.S.A.), similar to previously reported (Beaulieu 2007). Slice thickness was measured with an electronic digital caliper. Uniform “industry standard” cubes (approximately  $2.5 \times 2.5 \times 2.5$  cm) were

prepared, which generally served as a “control” tissue sample. Thin-sliced tissue and cubes were prepared from full-slip “Esteem” fruit, and exposed to an open atmosphere for 15 min to mimic handling and/or treatment time, followed by 1 and 3 d storage at 5 °C. To assess volatile retention in relation to possible boundary layer effects, simulated washing/rinsing exposures were induced by 1 min treatments in deionized water at 5 °C. Further sub-sampling of roughly 1 to 2 mm thick “external” surface tissue was excised to evaluate compounds in the boundary layer, compared with 1 cm<sup>3</sup> of previously undisturbed tissue excised from the “internal” cube center. “Washed” cube samples and thin-sliced tissue was stored at 5 °C up to 3 d, and cubes were stored at 5 °C in Juice Catcher clamshell containers (Pactiv Corp., Lake Forest, Ill., U.S.A.) for up to 9 d.

### Physiological measures

Percent soluble solids were measured from extracted juice or expressed cubes with a hand held electronic refractometer (PR101, Atago, Tokyo, Japan). Firmness was not measured in thin-sliced samples since it was not possible to measure excessively thin sliced tissue, and SEM was therefore utilized. Color ( $L^*$ ,  $a^*$ , and  $b^*$ ) was recorded with a Hunter color meter (DP-9000, D25A, Hunter Assoc. Lab. Inc., Reston, Va., U.S.A.) calibrated against a white (93.088, −0.689, −0.099, for  $L^*$ ,  $a^*$ ,  $b^*$ , respectively) color tile. Color readings were taken from the sides of each sample that were sliced with sharp knives, not the soft internal cavity-side or the external side peeled by the Muro Peeler. Weight loss (water, g) was determined by weighing tared samples periodically on an analytical balance.

### Surface area to unit volume measurements

Typical thin slices prepared throughout this study were roughly  $4 \times 3 \times 0.2$  cm. Within the industry, cubes are commonly cut at approximately 2.5 cm<sup>3</sup>. Thin slices and cubes from the equatorial region of 3 cantaloupe were prepared and measured ( $n = 9$ ) with an electronic caliper.

### Scanning electron microscopy (SEM)

Analytical texture evaluations were not practical due to the nature of thin-sliced cantaloupe tissue. Therefore, the exposed surface area of fresh or stored cantaloupe was imaged via SEM after various cutting and storage treatments to assess physical damage. Surface images were acquired on thin-sliced cantaloupe (store bought cultivar 1, ripe) prepared with the mandolin slicer, and fresh-cut cubes (approximately 2.5 cm<sup>3</sup>). A v-shaped wedge was razor-cut on the top (upside) surface of fresh-cut tissue to demark the proper surface orientation for imaging. Fresh samples were fixed directly into 3% glutaraldehyde in 0.05 M sodium cacodylate buffer, pH 7.4, for a minimum of 48 h at room temperature. After fixation, samples were washed in several changes of 0.05 M sodium cacodylate buffer and then dehydrated in a graded series of ethanol, up to 100%, over several days. After 2 changes in 100% ethanol, 1 slice of each sample was placed in liquid nitrogen and cryofractured (“cryo” mount). These portions were returned to 80% ethanol solution and then increased to 100% ethanol several times. Remaining samples were critical point dried from ethanol in liquid carbon dioxide and mounted on standard  $\frac{1}{2}$  inch SEM mounts using double-sided adhesive tabs. Samples were coated in a Hummer II Technics Coater (Alexandria, Va., U.S.A.) with gold/palladium (60%/40%) for 2 min (@ 10 V, 10 to 15 milliamps). Images were taken in conventional SEM mode on a FEI XL30 ESEM (FEI Inc. Phillips, Hillsboro, Oreg., U.S.A.) using

Polaroid T55 p/n film, and saved as tif files. Imaging was completed to avoid displaying surfaces where sample preparation or tissue mounting led to cracking, pitting or surface distortion. Cell diameters (avoiding vascular regions) were calculated by measuring 15 representative cells twice (perpendicular, width, and height) per micrograph, calculated against the 200 or 500  $\mu\text{m}$  bar.

### SPME GC-MS volatiles analysis

The sampling and GC-MS method described by (Lamikanra and others 2002) was employed for thin slices, with minor modifications (Beaulieu 2007). Both thin slices and fresh-cut cubes were analyzed as well, as previously described (Beaulieu and Grimm 2001; Beaulieu 2006b). Roughly 3 g macerated slices or cubes was placed in 10-mL vials with 1 g NaCl, 100 ppb (w/w, final concentration) amyl isovalerate internal standards (IS), magnetic stir bar, and capped with a Teflon/silicone septum. Samples were placed on a Combi-Pal (Leap Technologies, Carrboro, N.C., U.S.A.) autosampler at 30 °C for 15 min agitation followed by 15 min SPME adsorption period. Solid phase microextraction was employed using automated 1-cm 100- $\mu\text{m}$  PDMS fibers (Supelco Inc., Bellefonte, Pa., U.S.A.) and volatile compounds were thermally desorbed into the injection port of an Agilent 6890 GC for 4 min at 270 °C, containing a Zebtron ZB-5 (cross-linked 5% phenyl methyl silicone (Phenomenex, Torrance, Calif., U.S.A.) column (30 m, 0.25 mm i.d., 1  $\mu\text{m}$  film thickness) coupled to an Agilent 5973 MS. Cryofocusing at -70 °C was employed during desorption and the GC oven was initially held 1 min at 50 °C in the splitless mode. The oven temperature was then increased at 5 °C  $\text{min}^{-1}$  to 100 °C, then 10 °C  $\text{min}^{-1}$  to 190 °C and 30 °C  $\text{min}^{-1}$  to 250 °C and held for 2.33 min. The quadrupole MS was operated in the electron ionization mode at 70 eV, with a continuous scan from  $m/z$  33 to 300. Samples were run in triplicate and peak areas based on selected unique ions were normalized on amyl isovalerate, and averaged. Representative data is presented from repeated analyses. Identification of single components was performed by comparison of mass spectra (NIST Database, version 1.5, Palisade Corp., Newfield, N.Y., U.S.A.), authentic standards, and an in-house RI based on n-alkanes (heptane – eicosane), as described elsewhere (Beaulieu 2006b, 2007). Relative percentage recovery of selected compounds was based on target ion abundances or calculated based on normalization via the amyl isovalerate internal standard (IS). Esters were differentiated and classified based on carbon number and size as low molecular weight (LMW; < C<sub>9</sub>, not including benzyl acetate) against high molecular weight (HMW), since loss of esters resulting from UV treatments decreased markedly at > C<sub>8</sub> molecular weights (Beaulieu 2007). Total of 18 esters were extracted out of the volatile profile based on unique target ion responses, and were summed, normalized on the IS, per replicate, then averaged. Total of 11 LMW compounds (ethyl acetate, ethyl propanoate, propyl acetate, butyl acetate, ethyl 2-methylpropanoate, methyl 2-methylbutanoate, ethyl butanoate, ethyl 2-methylbutanoate, 2-methylbutyl acetate, 3-methylbutyl acetate, and 2-methylpropyl acetate), and 7 HMW compounds (ethyl hexanoate, hexyl acetate, benzyl acetate, ethyl benzoate, ethyl-2-phenyl acetate, 3-phenylpropyl acetate, and 2-phenylethyl acetate) were assessed. In all volatiles reported, only results using the most mature fruit were presented since the profiles were considered “complete,” unlike several representative repeats performed with store bought cultivar nr 2, and less ripe “Esteem” tissue, even though the trends remained constant.

## Results and Discussion

As volatile loss in UV-treated thin-sliced cantaloupe appeared to be an artifact (Beaulieu 2007), tissue sampling, washing protocol, physiological changes, surface appearance (SEM), GC-MS regimes, and volatile recovery in cut and treated melons were further examined. The purpose of analyzing thin slices was to maximize theoretical germicidal or surface area rinsing effects if aggressive food safety sanitation treatments were implemented.

### Physiological differences: Color, appearance, and translucency

Producing 1 mm thick cantaloupe slices on a Hobart slicer was difficult, as they generally tore and were immediately compromised, appearing water-soaked and translucent. Preparation of 2 mm thick slices ( $1.94 \pm 0.08$  mm,  $n = 18$ ) was more feasible, as the tissue remained intact and did not have poor visual appearance. However, a hand-held plastic kitchen slicer (mandolin) with a straight-edge blade producing from approximately  $2.31 \pm 0.16$  to  $2.67 \pm 0.11$  mm thick ( $n = 40$ ) slices with superior visual appearance was preferred. Slices did have a somewhat nonuniform thickness (from top to bottom) yet, slices were not translucent appearing. Damaged or nonuniform (thickness) sections were trimmed and discarded. Total 3 mm slices produced on the Hobart had the best visual appearance and they also maintained better appearance during storage, with less water-soaking, as observed in color trends reported subsequently. However, few slices could be produced per fruit using only equatorial tissue, and subsequently, approximately 2.5 mm slices produced with the mandolin slicer were generally utilized.

After 15 min, thin-sliced (1 and 2 mm) tissue already lost the normal wet gleam, and appeared drier. After 1 h thin-sliced samples were profoundly different looking; literally dried out on the surface due to desiccation, and 1 and 2 mm thin slices were extremely translucent. Translucency is commonly observed in severely wounded tissue (Brecht and others 2004). The  $L^*$  color change in tissue slices against standard cantaloupe cubes also indicated quality deterioration (Figure 1). A 4.1% to 12.6% decrease in  $L^*$  occurred immediately due to cutting (time 0), and after 3 d storage, thin-sliced tissue had  $L^*$  values that were 27.5%, 40.5%, and 52.9% lower for 3, 2, and 1 mm thickness,

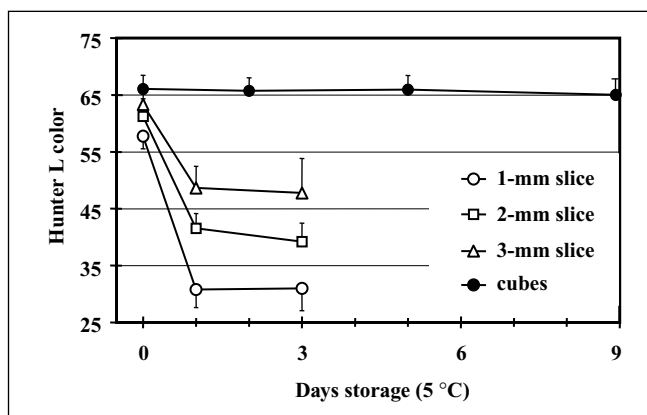


Figure 1—Comparison of Hunter ( $L^*$ ) color change in typical fresh-cut cubes (“Sol Real”) and thin-sliced “Esteem” cantaloupe tissue (approximately 1, 2, and 3 mm) stored at 4 and 5 °C, respectively. Cube data from 2 y composite sampling ( $n = 120$ ) whereas thin-sliced data is one representative trial of a repeated analysis ( $n = 9$ ), standard deviation indicated per data point.

respectively. It has been demonstrated that commercial-like fresh-cut cantaloupe cubes suffer minimal short-term color loss under optimum storage conditions (Portela and Cantwell 2001; Boynton and others 2005; Beaulieu and Lea 2007), and this was confirmed herein. Thin-sliced tissue was analyzed only 3 to 4 d due to severe physical and physiological quality reduction. Overall color (C) decreased in freshly prepared cubes (2.4%) and slices (14.4%) that were washed in cold water (Figure 2). Again, thin-sliced tissue had substantial overall color reduction (17.4%) on day 0, compared with cubes, and this trend continued throughout storage. Greater error bars observed in thin-sliced samples may indicate nonuniform translucency.

### Weight loss and soluble solids change

In thin-sliced tissue exposed to air for 15 min at 5 °C (as might occur during experimental preparation, handling, washing, or subsequent irradiation treatments and packaging), there was 3.3%, 1.8%, and 1.2% water loss after 15 min in 1, 2, and 3-mm tissue, respectively (Figure 3). After simulated abuse/treatment exposure, tissue was placed into 5 °C storage in static dishes, and weight loss was monitored. Substantial weight loss continued after 1 h, with 8.8%, 3.4%, and 2.8%, in 1, 2, and 3-mm thin-sliced tissue, respectively. The following day there was 20.4%, 9.5%, and 6.7% weight loss in 1, 2, and 3-mm slices, respectively (Figure 3). Cutting effectively exposes large cells of spongy parenchyma tissue that lose water readily to the drier atmosphere. These data indicate extensive water loss occurred in thin-sliced tissue, which was obviously driven by the water potential gradient (Nobel 1991), as subsequent data corroborate.

Soluble solids (°Brix) change was evaluated as another index to measure possible physiological alterations due to sampling and cutting treatments. Average soluble solids content of cubes (store bought, cultivar nr 1) was  $9.4\% \pm 0.9\%$  (SD) and this decreased 15.4%, to  $8.0\% \pm 0.5\%$ , upon immediate washing. Almost identical soluble solids losses were observed in thin-sliced tissue whereby unwashed controls had  $9.1\% \pm 0.4\%$  which dropped 15.8%, to  $7.6\% \pm 0.7\%$  upon rinsing. These data indicate that substantial soluble solids loss occurred in response to rinsing, independent of tissue cutting method. However, calculating percent soluble solids loss per averaged surface area

(see subsequently) indicates that thin slices essentially lost 22.4% soluble solids based on exposed surface areas, compared with cubes.

### Surface area to unit volume (SA : Vol)

Measurements from three typical fruit indicated that the surface area of thin-sliced tissue (approximately  $4 \times 3 \times 0.2$  cm) was roughly  $26.8 \pm 1.0$  cm<sup>2</sup> compared with  $38.0 \pm 1.5$  cm<sup>2</sup> for industry-style cubes (approximately 2.5 cm<sup>3</sup>). Slices used for this calculation were prepared with a Mandolin slicer (thickness,  $2.31 \pm 0.16$  mm). Of critical importance was the surface area (cm<sup>2</sup>) per unit volume (cm<sup>3</sup>) for thin slices ( $9.86 \pm 0.60$ , cm<sup>2</sup> : cm<sup>3</sup>) compared with  $2.39 \pm 0.05$  for cubes. Surface area per unit volume (SA : vol) in slices was 4.1 times greater than a typical fresh-cut cube. According to the previously published method where 1-mm thick slices of tissue were utilized (Lamikanra and others 2002; Lamikanra and others 2003; Lamikanra and Richard 2002), the SA : vol would be 8.9-fold greater than a typical cube. Therefore, a much greater water potential gradient exists in thin slices compared with cubes (Nobel 1991). Extensive weight loss occurred in thin-sliced tissue (Figure 3), and similar weight loss (or juice loss) is seldom evident in standard fresh-cut cantaloupe cubes, even after extended storage (data not shown). Increasing tissue surface area via thin-slicing, or even reducing cube size to maximize sanitation effect (adsorption, coverage, or penetration) leads to rapid deterioration compared with standard cubes.

### Scanning electron microscopy (SEM) appraisals

Upon cutting, the average cell diameter in thin-sliced cantaloupe was  $159.2 \pm 34.0$  μm (Figure 4A), and cell diameter of cells in a typical cube were  $148.1 \pm 41.8$  μm (Figure 4B). Care was taken to avoid measuring regions containing vascular bundles, as illustrated in a magnified (800×, 20 μm) panel, Figure 4C. Cell diameter decreased markedly (65.4%) to  $55.1 \pm 25.8$  μm in thin slices stored 1-d in static (sealed) Petri dishes at 5 °C (Figure 5A). However, cell diameter was still roughly  $122.6 \pm 51.2$  μm in 1-d old fresh-cut cubes stored at 5 °C, even though slight surface dehydration and cell wall peeling was apparent (Figure 5B).

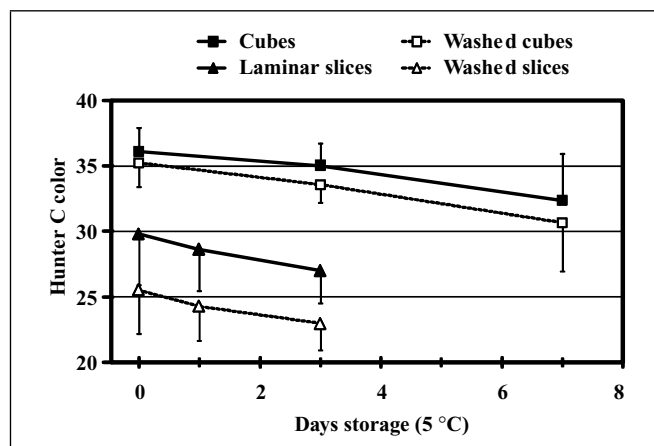


Figure 2—Hunter color (C) change in washed (1 min deionized water bath at 5 °C) and unwashed store-bought (cultivar 1) cantaloupe cubes and thin-sliced tissue (approximately 2.6 mm) stored at 5 °C in juice catcher containers. All data from equatorial mesocarp tissue of 4 replicate fruits, sampled 9 times each,  $n = 36$ , standard deviation indicated per data point.

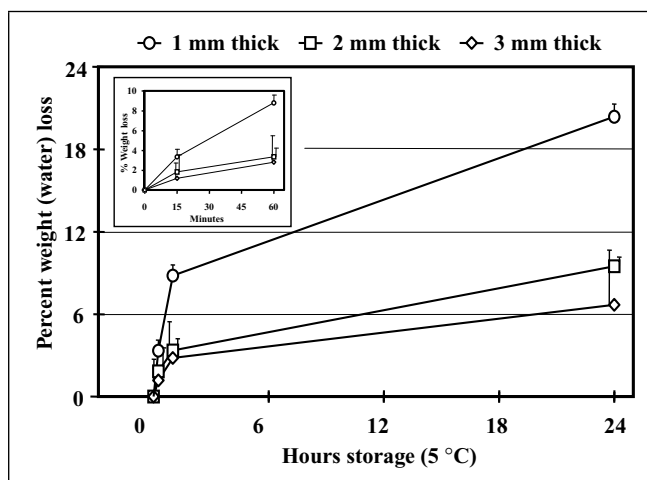


Figure 3—Water loss (percent weight) in thin-sliced ripe "Esteem" cantaloupe tissue (approximately 1, 2, and 3 mm thickness) exposed to room conditions for 15 min, then stored at 5 °C for 24 h. All data from a representative repeat using equatorial mesocarp tissue of 3 similar replicate fruits, sampled 3 times each,  $n = 9$ . Standard deviation indicated per data point (overlapping error bars moved to right on 2 mm squares, and left on 3 mm diamonds).



Cell size ( $83.6 \pm 28.3 \mu\text{m}$ ), and more importantly, structure, were still somewhat conserved after 4 d in fresh-cut cubes (Figure 5C). As comparison to 1-d-old thin-slices, cell size in fresh-cut cubes decreased only 43.5%, after 4 d storage in clamshell containers at 5 °C. Aside from slight and general cell size decrease observed in numerous 1- and 4-d-old cube micrographs, both had very similar appearances. This indicated that physical changes and possible desiccation and associated water loss were inconsequential, compared with thin-sliced tissue. No SEM images were acquired on thin-sliced tissue after 3 d since they were either decayed or

severely desiccated. In general, stored slices (24 h) had dramatically reduced cell size, dehydrated and/or folded cell walls, and apparent compression and/or tissue separation. These thin-sliced results are similar in nature to SEM reports indicating severe tissue desiccation, cell sloughing, and physical consequences observed in storage when carrot tissue was sliced with dull apparatus (Tatsumi and others 1991; Barry-Ryan and O'Beirne 1998). Our data visualize clearly that maximizing tissue surface area for possible efficacious food safety or irradiation treatments resulted in unacceptable quality loss.

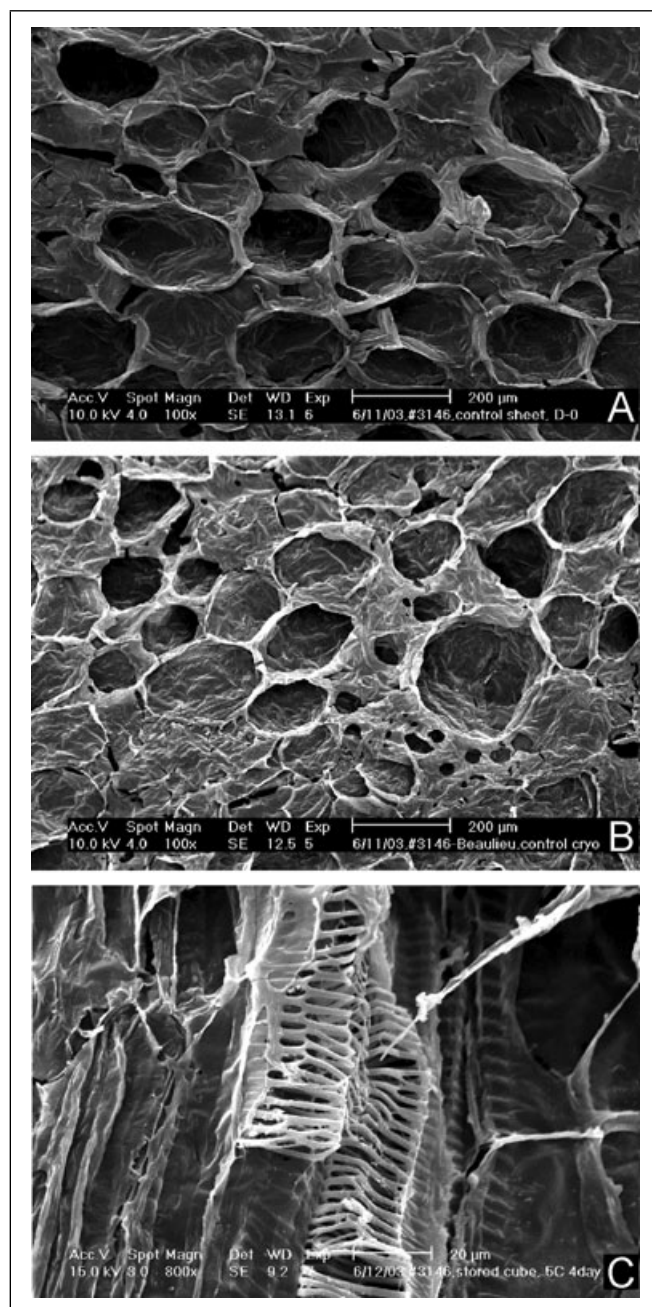


Figure 4—Surface images of fresh and stored fresh-cut cantaloupe (store bought, cultivar 1) via scanning electron microscopy. Typical images (100x, 200  $\mu\text{m}$ ) are presented for a normal SEM mount of freshly cut, thin-sliced (approximately 2.5 mm) cantaloupe (A), a cryofocused mount of freshly cut cantaloupe cubes (B), and vascular bundles in cut cubes, magnified (800x, 20  $\mu\text{m}$ ) in panel (C).

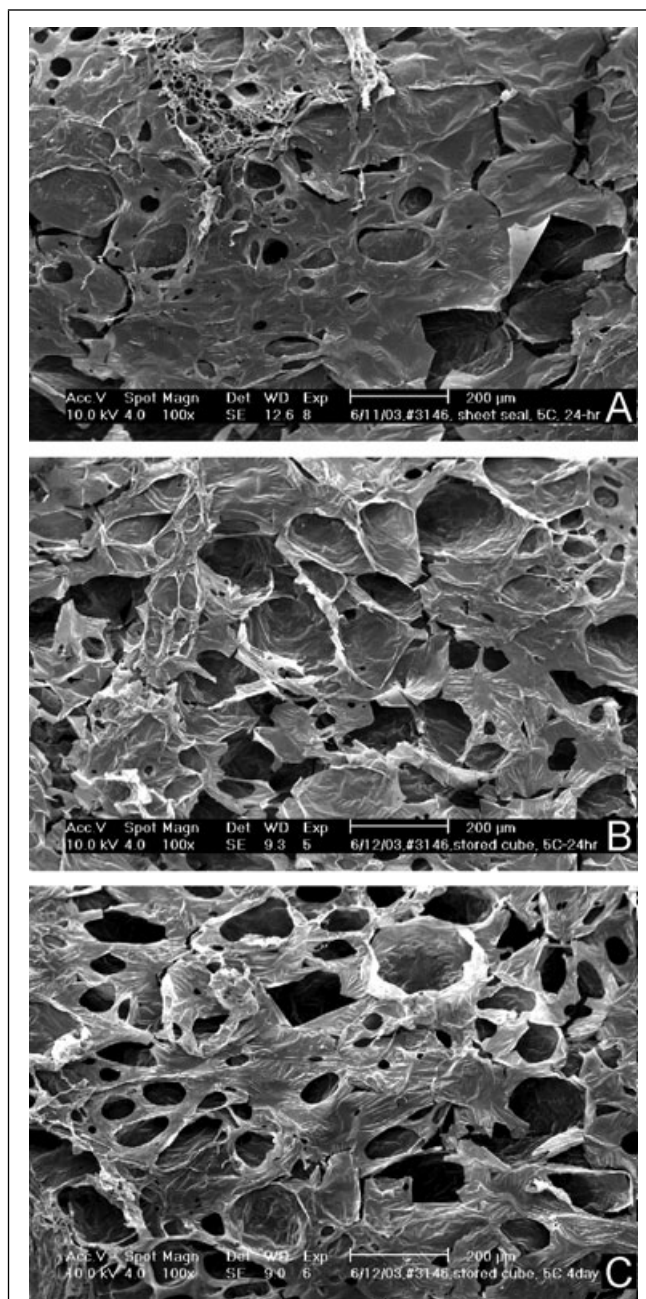


Figure 5—Surface images (100x, 200  $\mu\text{m}$ ) with normal SEM mounts of typical approximately 2.5 mm thin-sliced cantaloupe (store bought, cultivar 1) stored at 5 °C for 1-d in sealed containers (A), compared with cubes stored in juice catcher containers for 1 (B) and 4 (C) d.

## Volatile changes resulting from cutting and sampling regimes

There was substantial ester loss only in thin-sliced tissue, compared with cubes. Total esters decreased 92.8% and 95.8% after 1 and 3 d storage in sealed containers, respectively (Table 1). Like previously reported ester losses (Beaulieu 2007), the decline of HMW esters was appreciably lower (65.7%) after 1-d storage compared with LMW esters (96.7%). After 3 d storage, there was 95.8% loss of the 18 esters in thin-sliced tissue. On the other hand, total esters tended to increase in stored cubes that remained in sealed clamshell containers, independent of sampling method. Transient volatile increases followed by a general decline have been observed in numerous fresh-cut fruits (Beaulieu and Lea 2003a, 2003b; Beaulieu 2005, 2006a, 2006b; Saftner and others 2006), and in sensory aroma/flavor attributes (Bett and others 2001; Bett-Garber and others 2003). One exception to the trend was observed in a decline in HMW esters in cubes which were sub-sampled as thin-sliced tissue. We attribute this to cutting/handling tissue twice, and improper integration resulting from RT shifts and peak lowering when the GC-MS cryofocussing failed in some of these samples.

Cubes were sampled in an identical manner as the thin-sliced tissue, or homogenized into slurry, as customarily performed in our lab. A slight reduction in ester recovery in cubes seems a likely consequence resulting from technically handling samples twice while extracting volatiles, and slight oxidative loss that can occur during blending. Abusive (1 min) blending was shown to reduce esters and increase oxidatively generated compounds (Wyllie and others 1994); however, the blending protocol used herein was only 15 s, and aldehydes did not increase in untreated thin-sliced cantaloupe, while terpenoid oxidation was often evident in UV-treated tissue (Beaulieu 2007). These trends are reflected by similar ester profiles in both cube sampling regimes for selected compounds (Figure 6). Again, ester levels were similar on day 0 for all sampling regimes. However substantial ester decreases were observed in thin-sliced tissue after 1 and 3 d, as illustrated for 2 typical flavor-related compounds, ethyl hexanoate (Figure 6A) and ethyl 2-methyl butanoate (Figure 6B). As further evidence of tissue abuse, if thin-sliced tissue was prepared and sampled as Braun slurries (typical method in our lab), there was a massive decrease in ester recovery (91.3% HMW and 83.5% total) immediately after preparation on day 0 (Table 1). It has been reported that homog-

enizing the flesh of cantaloupe activates lipoxygenase (LOX) and hydroperoxide lyase (Wyllie and others 1994; Lamikanra 2002), even though these enzymes were not measured in these reports. However, 1 melon studied was green fleshed (Wyllie and others 1994) and there is LOX activity in the mesocarp (edible portion) of green fleshed honeydew melons (Lester 1998) whereas, there is little to no LOX activity in the mesocarp of certain orange-fleshed cantaloupe (Lester 1990).

Washing tissue provided an apparent boundary layer or film that encapsulated volatiles on day 0, and for up to 3 d in both

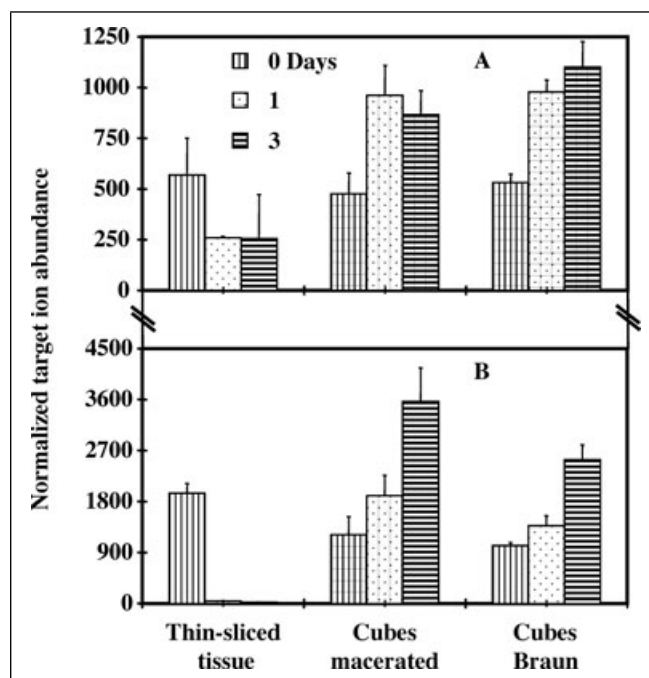


Figure 6—Normalized GC-MS response for typical flavor-related compounds in stored (5 °C) thin-sliced "Esteem" cantaloupe tissue (approximately 2.6 mm) and cubes. LMW ethyl hexanoate (A) and ethyl 2-methyl butanoate (B). Representative repeat displayed for  $\frac{3}{4}$ -slip harvest, ripe fruit,  $n = 3$ , standard deviation indicated per data point.

**Table 1—Normalized GC-MS SPME ester recovery in fully-ripe "Esteem" cantaloupe sampled as thin-sliced tissue (approximately 2.6 mm) that were exposed to an open atmosphere at ambient temperature for 15 min, compared with fresh-cut cubes, held identically, then stored at 5 °C.**

	Days storage		
	0	1	3
Thin-sliced tissue			
LMW compounds <sup>a</sup>	21,491 ± 4,414	707 ± 105	757 ± 288
HMW compounds	3,135 ± 1,014	1,076 ± 196	269 ± 219
Total	24,626 ± 5,428	1,782 ± 302	1,027 ± 508
Cubes, sampled as thin-slices			
LMW compounds	15,485 ± 3,321	22,399 ± 3,322	30,555 ± 5,008
HMW compounds	2,995 ± 697	3,599 ± 409	1,397 ± 150
Total	18,480 ± 4,018	25,997 ± 3,730	31,952 ± 5,159
Cubes, Braun slurry			
LMW compounds	11,028 ± 561	14,456 ± 1,426	18,478 ± 2,291
HMW compounds	1,293 ± 196	2,533 ± 474	1,465 ± 175
Total	12,321 ± 757	16,989 ± 1,899	19,943 ± 2,465
Thin-slice, Braun slurry			
LMW compounds	3,799 ± 966	—	—
HMW compounds	274 ± 59	—	—
Total	4,073 ± 1,025	—	—

<sup>a</sup>Total of 11 LMW compounds and 7 HMW compounds were analyzed according to the materials and methods. All data from a representative repeat using equatorial mesocarp tissue of 3 similar replicate fruits, standard deviation indicated.

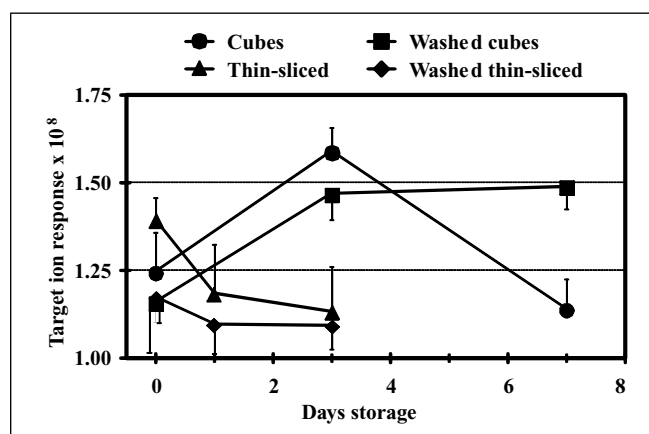


Figure 7—Volatile recovery of 18 quantified esters (GC-MS target ion response) immediately post-cutting and after sealed storage at 5 °C in wetted thin-sliced (approximately 2.6 mm) and cantaloupe cubes (store bought cultivar 1) that were washed for 1 min in deionized water at 5 °C. Representative repeat displayed for ripe fruit,  $n = 4$ . Standard deviation indicated per data point (overlapping error bars moved to right on washed cube squares, and left on washed thin-sliced diamonds).



thin-sliced tissue and cubes (Figure 7). Edible coatings were likewise responsible for entrapment of flavor volatiles in fresh-cut apples (Bai and others 2002; Olivas and others 2007). These results may indicate that like edible coatings, simply washing tissue may provide a sufficient short-term wetting to reduce volatile losses. Again, there was a curious transient increase in control cubes, followed by decline (day 7), and as previously noted, transient volatile increases are not uncommon in fresh-cut fruits. Day 7 volatiles were not analyzed in thin-sliced tissue since they had deteriorated substantially (desiccation, decay, and/or translucency). The volume of a thin slice likely limits available water/solute to replace that driven off at the surface, leads to rapid desiccation, and likely limits the available pool of substrates necessary to metabolize or catabolize and deliver additional volatile compounds (Beaulieu 2006a) to the surface, over time.

Since there were marked differences in volatiles recovered from stored thin-sliced tissue against cubes, an appraisal of tissue was made to assess volatile differences in external surfaces versus internal tissue. Thin-sliced tissue that was water dipped (deionized washed) at 5 °C for 1 min and stored at 5 °C for 0, 1, and 3 d had an extremely similar volatile profile on day 0 compared with internal and external tissue (1–2 mm edges) excised from cubes prepared from the same equatorial and adjacent tissue (Figure 8). These data provide a clear indication that a somewhat stable short-term volatile profile is maintained from the surface of a cube with an appreciable volume, as compared with the interior tissue of that same cube. There was a substantial ester loss after 1 and 3 d storage (50.1% and 61.7%, respectively) that only occurred in washed thin-slices. Comparing the washed tissue to unwashed samples (Table 1, thin-slices with total ester decreases of 92.8% and 95.8% after 1 and 3 d storage, respectively), provides a clear indication that ester loss was not as marked when the surface of exposed tissue was wetted (Figure 8). These data indicate that thin-sliced tissue rapidly lost important volatile esters; however, once again, the cubes maintained volatiles significantly longer, especially the sub-sampled internal tissue. There are also likely differences in volatiles recovered from washed cubes against control cubes (Figure 7) due to the water vapor effectively forming a boundary layer against volatilization.

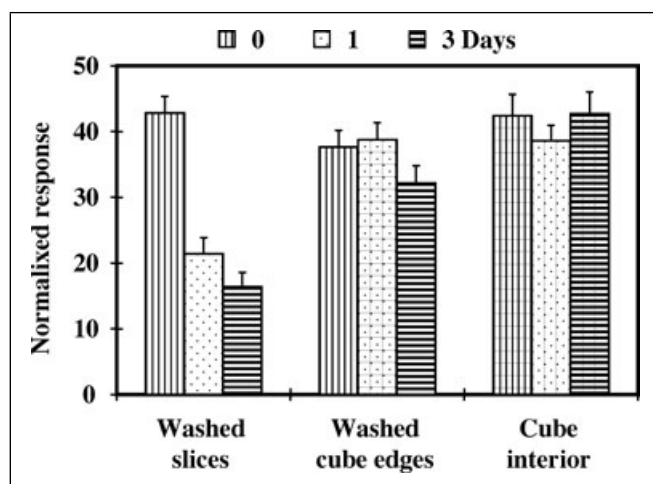


Figure 8—Boundary layer effects assessed by GC-MS volatile recovery (normalized response, 18 esters) in surfaces of washed (1 min, deionized water at 5 °C) thin-sliced “Esteem” cantaloupe ( $\frac{3}{8}$ -slip harvested) and cubes, compared with internally extracted cube tissue. Representative repeat displayed for ripe fruit,  $n = 4$ , standard deviation indicated per data point.

## Conclusions

Preparing thin-sliced cantaloupe tissue resulted in marked color loss and tissue translucency, independent of whether or not tissue was rinsed or washed, compared to control cubes. The thin-sliced tissue also had substantial water loss after only 1 d in 5 °C storage. Rinsing both cantaloupe cubes and thin-sliced tissue resulted in roughly 15% soluble solids loss. SEM indicated that desiccation and water loss dramatically reduced cell size in thin-sliced cantaloupe tissue, resulting in excessive cell sloughing and damage, as compared with stored fresh-cut cubes. Results as such should be taken into consideration when sanitation rinses are employed in fresh-cut fruits.

Thin-sliced cantaloupe tissue had substantially lower volatile esters recovered compared with fresh-cut cubes during storage. LMW esters were lost in greater quantities compared with HMW esters. LMW esters will be more rapidly off-gassed due to water potential gradients, low boiling temperature, high flash point (vapor pressure), and surface area to volume relationships. Cantaloupe tissue samples exposed to UV in a closed static system did not have dramatic ester losses as compared with tissue treated in an open ambient system (Beaulieu 2007). Herein, washing cantaloupe tissue effectively also provided a short-term barrier to volatile loss in both cubes and thin-sliced tissue. Nonetheless, cutting method and sampling protocol were determined to be responsible for volatile losses, leading to possible misinterpretation of flavor attributes and quality in previously reported cut cantaloupe (Lamikanra and Richard 2002; 2004; Lamikanra and others 2003). For example, dramatic cutting and static storage in papaya was shown to decrease respiration and alter membrane degradation, free radical generation, and global stress response enzyme activities (Karakurt and Huber 2007). Since volatile recovery has been demonstrated to be clearly compromised when tissue is severely damaged (thinly cut) against standard industry cutting (for example, cubes), it is rational to assume that consequent physiological deterioration and physical tissue abuse likewise compromised normal enzyme activities. In light of sample preparation-dependent quality and ester loss reported herein, we therefore disagree with the conclusions and validity of several volatile and enzymatic studies emanating from previous experiments using the thin-sliced methodology.

The current data demonstrates that cutting method and sampling protocol are critically important when using volatile recovery as a means by which to assess or interpret stress response and ascribe fresh-cut quality. In somewhat less firm fresh-cut fruit with large cell sizes (for example, melon, papaya, pear, and watermelon), there is a trade-off between possible benefits attained by washing and rinsing treatments geared toward microbial reduction or tissue firming. These treatments may provide a protective boundary layer that entraps volatiles yet, soluble solid losses occur during rinsing. Likewise, reduction of tissue size to maximize food-safety sanitation efficacy (for example, thin-sliced) or deliver items to a niche market (for example, small-cut cubes for children) will need substantial work to engineer equipment and protocols to insure product quality and volatile profiles are not compromised. These relationships should be explored in future sensory and consumer evaluations.

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